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Published in:
Genome Announcements

DOI:
[10.1128/genomeA.01108-15](https://doi.org/10.1128/genomeA.01108-15)

Publication date:
2015

Document version
Publisher's PDF, also known as Version of record

Citation for published version (APA):
Dolka, B., Boyen, F., Butaye, P., Olsen, R. H., Thøfner, I., & Christensen, J. P. (2015). Draft genome sequences of two commensal *Enterococcus cecorum* strains isolated from chickens in Belgium. *Genome Announcements*, 3(5), [e01108-15]. <https://doi.org/10.1128/genomeA.01108-15>

Draft Genome Sequences of Two Commensal *Enterococcus cecorum* Strains Isolated from Chickens in Belgium

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Here, we report the draft genome sequences of two commensal *Enterococcus cecorum* strains (1710s23 and 1711s24), cultivated from the ceca of healthy laying hens originating from different farms in Belgium.

Received 11 August 2015 **Accepted** 17 August 2015 **Published** 24 September 2015

Citation Dolka B, Boyen F, Butaye P, Heidemann Olsen R, Naundrup Thøfner IC, Christensen JP. 2015. Draft genome sequences of two commensal *Enterococcus cecorum* strains isolated from chickens in Belgium. *Genome Announc* 3(5):e01108-15. doi:10.1128/genomeA.01108-15.

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Enterococcus cecorum is a normal inhabitant of the gastrointestinal flora of mammals and birds (1). It is the most frequently occurring enterococcal species in the intestine of adult chickens (2). Recently, several *E. cecorum*-related clinical cases in poultry have been reported worldwide (3–6).

Here, we report two draft genome sequences of commensal *E. cecorum* strains that were isolated from the ceca of 2 laying hens from different farms in Belgium. The chickens were submitted in August 2009 to the Department of Pathology, Bacteriology, and Avian Diseases, University of Ghent. Birds did not show clinical signs or necropsy findings associated with disease. *E. cecorum* was not isolated from any of the extraintestinal organs. For each bird, cecal content was collected aseptically and plated on sheep blood agar supplemented with colistin and nalidixic acid (Biomedix Ltd., Canada) and incubated for 24 to 48 h at 35°C in 5% CO₂. Isolates were recovered from one bird per farm. The identification was performed by API 20S (bioMérieux, France) and further confirmed by sequencing of the 16S rRNA genes (7) and alignment with the sequences of the reference strains. The recovered *E. cecorum* isolates were assigned as control (intestinal) isolates and used for comparison studies (8). PFGE (Pulsed Field Gel Electrophoresis) revealed a large genetic diversity of *E. cecorum* from noninfected control chickens (8).

Genomic DNA of *E. cecorum* strains 1710s23 and 1711s24 was extracted using the DNeasy blood and tissue kit (Qiagen, USA). DNA quantity and quality were assessed using Nanodrop spectroscopy (Thermo Scientific, USA). Genomes were sequenced by the Illumina paired-end method (MiSeq 150) using a paired-end library with an average read length of 2 × 150 bp. Reads were *de novo* assembled using the CLC Genomics Workbench version 7.0. The draft genome assembly of 1710s23 is 2.26 Mb, comprising 28 contigs (range, 271 to 424,975 bp), with an average length (N_{50}) of 170,910 bp and 36.4% G+C content. Similarly, the 1711s24 genome has 28 contigs (range, 1,407 to 424,966 bp), with a total genome size of 2.30 Mb, an N_{50} of 170,921 bp, and 36.2% G+C content. Both genomes were submitted to Rapid Annotations using Subsystems Technology (RAST) (9) and the NCBI Prokaryotic

Genome Automatic Annotation Pipeline (PGAAP, http://www.ncbi.nlm.nih.gov/genome/annotation_prok) (10) for annotation. The 1710s23 genome is estimated to contain 2,247 genes (12 rRNAs, 48 tRNAs) and 2,099 expected protein-coding sequences (CDSs). The 1711s24 genome contains 2,295 genomic features consisting of 2,143 CDSs (predicted), 46 tRNAs, and 10 rRNAs. No virulence factors were found by using the VirulenceFinder version 1.4 server (11).

To the best of our knowledge, this is the first genome report of commensal *E. cecorum* isolated from commercial laying hens in Europe. The data reported here may be useful in the investigation of genome variability of *E. cecorum* and for future comparative genomic studies on the pathogenicity of *E. cecorum*.

Nucleotide sequence accession numbers. The whole-genome shotgun projects have been deposited at DDBJ/EMBL/GenBank under the accession numbers LDOW00000000 (1710s23) and LDOX00000000 (1711s24). The versions described in this paper are the first versions, LDOW01000000 and LDOX01000000, respectively.

ACKNOWLEDGMENTS

This project has received funding from the European Union's Seventh Framework Programme for research, technological development, and demonstration under grant agreement no. 613574.

The work was supported by the University of Copenhagen, Faculty of Health and Medical Sciences, Department of Veterinary Disease Biology, and Warsaw University of Life Sciences—SGGW, Faculty of Veterinary Medicine, Department of Pathology and Veterinary Diagnostics.

REFERENCES

- Devriese LA, Ceyssens K, Haesebrouck F. 1991. Characteristics of *Enterococcus cecorum* strains from the intestines of different animal species. *Lett Appl Microbiol* 12:137–139. <http://dx.doi.org/10.1111/j.1472-765X.1991.tb00524.x>.
- Devriese LA, Hommez J, Wijnels R, Haesebrouck F. 1991. Composition of the enterococcal and streptococcal intestinal flora of poultry. *J Appl Bacteriol* 71:46–50. <http://dx.doi.org/10.1111/j.1365-2672.1991.tb04661.x>.
- Aitchison H, Poolman P, Coetzer M, Griffiths C, Jacobs J, Meyer M,

- Bisschop S. 2014. Enterococcal-related vertebral osteoarthritis in South African broiler breeders: a case report. *J S Afr Vet Assoc* 85:1077. <http://dx.doi.org/10.4102/jsava.v85i1.1077>.
4. Makrai L, Nemes C, Simon A, Ivanics E, Dudás Z, Fodor L, Glávits R. 2011. Association of *Enterococcus cecorum* with vertebral osteomyelitis and spondylolisthesis in broiler parent chicks. *Acta Vet Hung* 59:11–21. <http://dx.doi.org/10.1556/AVet.59.2011.1.2>.
 5. Robbins KM, Suyemoto MM, Lyman RL, Martin MP, Barnes HJ, Borst LB. 2012. An outbreak and source investigation of enterococcal spondylitis in broilers caused by *Enterococcus cecorum*. *Avian Dis* 56:768–773. <http://dx.doi.org/10.1637/10253-052412-Case.1>.
 6. Szeleszczuk P, Dolka B, Żbikowski A, Dolka I, Peryga M. 2013. First case of enterococcal spondylitis in broiler chickens in Poland. *Med Weter* 69: 298–303. <http://medycynawet.edu.pl/index.php/component/content/article/277/4660-summary-med-weter-69-5-298-303-2013>.
 7. Cai H, Archambault M, Prescott JF. 2003. 16S ribosomal RNA sequence-based identification of veterinary clinical bacteria. *J Vet Diagn Invest* 15: 465–469. <http://dx.doi.org/10.1177/104063870301500511>.
 8. Boerlin P, Nicholson V, Brash M, Slavic D, Boyen F, Sanei B, Butaye P. 2012. Diversity of *Enterococcus cecorum* from chickens. *Vet Microbiol* 157: 405–411. <http://dx.doi.org/10.1016/j.vetmic.2012.01.001>.
 9. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. *BMC Genomics* 9:75. <http://dx.doi.org/10.1186/1471-2164-9-75>.
 10. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Ciufu S, Li W. 2013. Prokaryotic Genome Annotation Pipeline. In Beck J, Benson D, Coleman J, Hoepfner M, Johnson M, Maglott D, Mizrahi I, Morris R, Ostell J, Pruitt K, Rubinstein W, Sayers E, Sirotkin K, Tatusova T (ed), *The NCBI handbook*, 2nd ed. National Center for Biotechnology Information, Bethesda, MD.
 11. Joensen KG, Scheutz F, Lund O, Hasman H, Kaas RS, Nielsen EM, Aarestrup FM. 2014. Real-time whole-genome sequencing for routine typing, surveillance, and outbreak detection of verotoxigenic *Escherichia coli*. *J Clin Microbiol* 52:1501–1510. <http://dx.doi.org/10.1128/JCM.03617-13>.